EJFA Emirates Journal of Food and Agriculture

Emirates Journal of Food and Agriculture 36: 1–10
doi: 10.3897/ejfa.2024.125334

RESEARCH PAPER

Prestorage hot water treatments retard papaya fruit deterioration and enhanced fruit quality in cold storage

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Academic editor: Siraj Beshir Sheko ♦ **Received** 13 April 2024 ♦ **Accepted** 22 June 2024 ♦ **Published** 31 October 2024

Abstract

Papaya (*Carica papaya*) fruit has a short shelf life and suffers significant postharvest losses due to climacteric, instant ripening and perishable nature. Pre-storage hot water treatment (HWT) has significant potential to delay decay incidence in horticultural commodities. The present study was designed to evaluate effectiveness of HWT on 'Red lady' papaya fruit for minimizing the fruit decay and maintaining the quality during storage. Physiologically mature unripe 'Red lady' papaya fruit harvested from a commercial papaya orchard. Selected fruits were treated at three different hot water temperatures (50 °C, 52 °C and 54 °C) along with a control (room temp) for 10 minutes thereafter stored at 12 °C and 85–90% RH for four weeks. Different sensory evaluation, fruit quality scales and enzymatic activities were observed at weekly intervals. Fruit stored after 54 °C HWT showed lower weight loss (4.63%), decay incidence (5.1%) and higher fruit firmness (12.92 N) relative to control. Fruit treated at 52 °C HWT improved the fruit acidity, but total soluble solids decreased than control. The results revealed that ascorbic acid (33.33 mg/100 g), total phenolic contents (35 mg/g FW) total antioxidants (47.50% DPPH inhibition), and antioxidative enzyme activities were improved by 52 °C HWT. It is concluded that pre storage papaya fruit treatment (HWT @ 52 °C) preserves the quality of fruit throughout storage on the other hand at 54 °C HWT helps in decreased fruit decaying. It can be summarized that papaya fruit can be treated with hot water in the range of 52–54 °C to support the fruit shelf life and suppress fruit decay along with maintaining fruit quality.

Practical applications: Papaya (*Carica papaya* L.) is a climacteric fruit which is highly susceptible to fungal attacks after harvest. Due to its perishable nature, it becomes decayed and huge amount of highly nutritious and medicinally important fruit is wasted. Recently people are more concerned about food safety which limits the use of fungicides for controlling the fruit decay in papaya. However, heat treatment has tremendous potential to control microbial decay, and keep food commodities safe from hazardous chemicals. In this study, the hot water treatment has successfully suppressed the fruit decay and maintained fruit quality of papaya in the course of entire period of cold storage. These findings indicated that use of hot water treatment is a practical approach for increasing the shelf life of papaya fruit.

Keywords

Antioxidative response, climacteric fruit, pre-storage treatment, physicochemical changes, shelf life

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Introduction

Papaya (*Carica papaya* L.) is an important fruit mostly grown in tropics and subtropics worldwide. Papaya is an excellent source of ascorbic acid, carbohydrates, fibers and vitamins with high amount of papain which aids in digestion (Oloyede 2005). Ripe papaya fruit has anti-aging, anti-cancer and laxative properties (Aravind et al. 2013). Despite papaya's high-water content (86–90%), it suffers postharvest losses which is reported 40–75% due to mechanical damage, dehydration, and pathogen attacks during postharvest processing (Al-Qurashi and Awad 2011). Postharvest losses of papaya during the whole marketing chain occur due to various reasons including pathological, physical or physiological disorders. Fruit softening and low temperature injury are the major constraints in long distance transport of Red lady papaya (Nunes et al. 2006). However, microbial decay, physical injury and loss of firmness are the major postharvest defects of papaya which limits its shelf life and shipment to distant places (Hernandez et al. 2006) due to poor eating and sensory quality.

Among all other attributes fruit decay and rotting is thought of as an important factor, when the term quality is discussed for the postharvest life of fruit. Papaya being climacteric and perishable may get affected by fungal decay, which deteriorates the postharvest quality of papaya fruit and becomes a hurdle in extension of its storability. Anthracnose (a fungal disease) is well known with reference to postharvest disease of papaya fruit (Cia et al. 2007) causing fruit decay. This highly nutritional fruit cannot reach distant places, because if it is transported under low temperature, it becomes susceptible to chilling injury and at high temperature ripening is triggered. Hodges et al. (2004) stated that chilling stresses disturb the balance among reactive oxygen species generation and immunity components. This equilibrium is precarious to cell endurance under cold storage span. Similarly for controlling fungal decay, different fungicides likewise propiconazole or prochloraz have been widely used as postharvest treatment. Nowadays consumers are more concerned for their health and discourage the use of fungicides as it has negative impact on human health due to fungicides and also the increase in environmental health issues (Gamage et al. 2004; Droby et al. 2009). It has been demonstrated that pesticide residues can raise the risk for cancer, endocrine disorders, and reproductive system disorders (Horrigan et al. 2002). It is estimated that there are about 385 million cases of acute unintentional pesticide poisoning each year, including 11,000 deaths; most of these cases occur in developing countries, including Pakistan (Boedeker et al. 2020). Hence, non-chemical methods are getting researchers attention to control microbial decay and maintain storage quality, including hot water treatment (Villa-Rojas et al. 2011), irradiation (Jouki and Khazaei 2014), etc.

Hot water treatment (HWT) prevents relatively huge number of postharvest losses occurred due to fungal problems, it is thought to be the most versatile, promising, effective and ecofriendly method (Mari et al. 2007; Jemric et al. 2011; Fruk et al. 2012; Liu et al. 2012). It has been long time to introduce HWT at commercial level as a postharvest treatment to control postharvest losses due to microbial pathogens (Schirra and D'Hallewin 1997; Hong et al. 2007; Liu et al. 2012), it also found effective in controlling chilling injury in several fruit crops (Schirra et al. 2005; Lu et al. 2010; Rui et al. 2010), inhibits fruit crop ripening, delay senescence and diminishes several physiological disorders (Fallik 2004; Ciou et al. 2011; Jemric et al. 2011), thus, helpful in maintaining quality of fruit and prolonging shelf life of horticultural crops. Heat stress is also the strategy for boosting the immune system of the plant to survive in adaptive environment and also inhibit chilling injury during storage (Lurie 1998; Fallik 2004).

In a nutshell, Hot water treatment can be an efficient strategy to restrain the incidence of fruit decay in friendly environment and maintain fruit quality by increasing the antioxidative enzyme activities. To the best of our knowledge, hot water application on 'Red lady' papaya has not been evaluated for pre-storage treatment, which becomes the strong base of this study. The objective of this study was to suppress the occurrence of decay and maintain the good quality of 'Red lady' papaya fruit under cold storage conditions.

Materials and methods

Fruit material, treatments and storage condition

Physiologically mature but unripe papaya fruit having traces (25%) of yellow colored skin were collected from a locally grown papaya orchard located at Faisalabad (30°51'41"N, 73°1'46"E), Punjab, Pakistan. The total numbers of fruits selected for the experiment were 260 that were sorted out on the base of size uniformity and disease free. The good quality, disease and insect free papaya fruit were wrapped in newspaper, packed in cardboard boxes and brought to the Research laboratory postharvest section, Ayyub Agriculture Research Institute, Faisalabad, Pakistan. Fruits were thoroughly washed with tap water for removing any soil or dust particles, then with chlorinated water and allowed to dry at room temperature (25+1 °C) for 1 hour. The fruits were divided into four groups and each group with four experimental treatments which were HWT @ 50 °C, HWT @ 52 °C, HWT @ 54 °C and control@ room temperature. All the fruit subjected to respective treatment for 10 minutes and then air dried under fan. Fruit treated with experimental application and controls were placed in plastic baskets to subject them for cold storage at 12+1 °C and 85–90% RH for 28 days.

Measurements of fruit weight loss (%), fruit firmness and fruit decay (%)

Three fruits from each experimental group were placed in net bag and weighed on weekly basis to observed weigh difference with respect to time. Fruits were weighed with

digital weighing balance (MJ-W176P, Panasonic Japan). Fruit weight loss was calculated by the following formula.

$$\text{Weight loss (\%)} = \frac{\text{Primary weight} - \text{Secondary weight}}{\text{Primary weight}} \times 100$$

Primary weight = weight at 0 day
Secondary weight = weight at weekly intervals

Papaya fruit was peeled off from two sides in opposite direction to insert the penetrometer (Model DFM50, Ametek Inc., USA) having 8 mm tip for measuring the fruit firmness expressed in "N". Fruit decay incidence is the visual inspection of decayed fruit (2–3 fungal or rotting spots) on daily basis to calculate the decay % at weekly basis decay %.

Total soluble solids (°Brix), titratable acidity (%) and ripening index

Fruit juice was extracted with juicer machine from fruit pulp and collected in beaker. A digital refrectrometer used for checking the amount of total soluble solids (Hanif et al. 2020). The amount of titratable acidity was assessed by standard method as outlined by Hortwitz (1960). For this purpose, papaya juice (5 ml) was mixed with distil water and poured into a conical flask (100 ml). Then the sample along with few drops of indicator (phenolphthalein) was titrated by 0.1 N NaOH till the persistence of end point (pink color). Titratable acidity was obtained by the formula:

$$\text{TA (\%)} = \frac{\text{0.1N NaOH used} \times 0.0064 \times 100}{\text{ml of Juice used}}$$

Ripening index was calculated in each sample by dividing the percentage of total soluble solids with the percentage of total titratable acidity.

Vitamin C, total phenolic and total antioxidants

The standard titration procedure of Ruck (1961) was adopted for measuring the ascorbic acid content of papaya juice sample for this purpose, extracted papaya juice (5 ml) was collected in a volumetric flask (100 ml) and extent was made by way of including 0.4% oxalic acid solution. Both ingredients were mixed and filtrated for collecting 5 ml aliquot sample which was titrated in opposition to 2, 6-dichlorophenol dye, to mild pink color stop point, persisted as a minimum for 15 seconds. The final vitamin C contents were expressed as mg/100 ml of juice.

The amount of total phenolic contents was measured by the procedure of Ainsworth and Gillespie (2007) as outlined by Hanif et al. (2020a). For this purpose, papaya juice (1 g) was mixed with a reagent (Folin-Ciocalteu) and reaction mixture. The acquired reading of spectrophotometer at 765 nm wavelength was compared with standard graph of blank sample and total phenolic contents were expressed as (mg GAE 100 g^{-1}). The total antioxidants were measured by using the procedure of Brand-Williams et al. (1995) as outlined by Hanif et al. (2020b).

Enzymes activity

Papaya juice 1 g in frozen form was homogenized using phosphate buffer and utilized as enzyme extract for estimating the CAT, SOD and POD activity. Enzyme extract was prepared by carrying out the procedure of Liu et al. (2009) for the estimation of catalase, peroxidase and superoxide dismutase activity (Hanif et al. 2020). The activity of catalase enzyme was checked by reacting the enzyme extract with hydrogen peroxide substrate. Changes in response were recorded as spectrophotometer reading, and activity was calculated as U/mg protein. The peroxidase activity was assessed by phosphate buffer 50 mM, H_2O_2 40 mM, and guaiacol 20 mM as a substrate (and recorded on spectrophotometer (470 nm). Superoxide dismutase activity calculated in in U/mg protein with substrate used distill water + 50 mM phosphate buffer + 20 mM NBT + 22 mM methionine + 0.1 mM Triton + 0.6 mM riboflavin.

Protein content for estimation of enzyme activities

Protein contents for the activity of antioxidative enzymes were taken out by Bradford method (Bradford 1976).

Statistical analysis

The experiment was set up as completely randomized design with two factors factorial arrangements for treatments with 4 replications. The collected data was analyzed by statistical software Statistix 8.1 for Analysis of variance and the means were compared by least significance difference test (LSD) (Steel et al. 1997). Graph and data presentation were performed with Microsoft office 2016.

Results

Fruit weight loss, fruit firmness and decay

Fruit weight loss is a continuous process due to metabolic activities of fruit and it increased with extended storage period in case of all treated and untreated fruit (Fig. 1a). Fruits with Hot Water treatments showed very little fruit weight loss reduction relative to control. Fruit weight loss showed significant variation among treatments and relative to control ($p \leq 0.05$). Papaya fruit under control conditions without HWT treatment showed highest fruit weight loss (9.75%) and very little weight loss (4.63%) in HWT @ 52 °C. Fruit weight decreased gradually from control to HWT @ 50 °C, HWT @ 52 °C and HWT @ 54 °C, respectively.

The fruit softening was much more notable in control fruit (from 19.58N to 10.70N) while least noticeable in

fruit exposed to HWT @ 54 °C (from 19.33°N to 12.91°N) on 28th day of storage. Fruit remained firm during 0–7 days of storage with negligible variations among all treatments. Untreated fruits were more rigid at 0 day while loses their rigidity and become soft at the end of storage. While fruit exposed to HWT @ 54 °C indicated high fruit firmness throughout observation in storage. At the end of experiment peak level of firmness was recorded in fruit exposed to HWT @ 54 °C accompanied by 52 °C, 50 °C and minimal was in control (Fig. 1c).

Fruit rotting showed significant variations among HWT and period of storage ($p \leq 0.05$). Fruits without HWT showed maximum fruit rotting (22.47%) followed by HWT @ 50 °C (15.51%), 52 °C (9.76%) and 54 °C (5.15%) (Fig. 1b). Fruit stored with control treated starts decaying in the first week to storage and reach up to the peak with abrupt increase in the 3rd week of storage. Fruit treated with HWT didn't indicate fruit rotting in the first two weeks. There was a positive interaction among storage span and rotting of fruit, as this span increases rotting also progressed.

Total soluble solids, titratable acidity and ripening index

Statistical analysis revealed differences in TSS accumulation over the course of storage time depending on HWT application, storage duration, and the relationship between the two. (Fig. 2a). Fruit with control treatment application exhibit lower TSS during first week while highest during last week of cold storage. Papaya fruit showed higher TSS in control followed by HWT @ 50 °C, HWT @ 54 °C and HWT @ 52 °C treatment. Untreated fruit depicted an uninterrupted rise in TSS level throughout storage; while HWT at 52 °C treated fruit revealed less accumulation (1.2-fold less than control fruit) in TSS in the course of whole storage.

Data being analyzed by Statistical software revealed that significant variations exist between HWT and storage span for acidity of juice. Fruit subjected to HWT @ 52 °C observed with higher acidity (0.05%) and minimum acidity level observed (0.043%) in control treatment (Fig. 2b). Fruit both exposed to lower temperature and untreated depicted alike response regarding TA with continuation of storage. Fruit exposed to HWT @ 52 °C and HWT @ 54 °C resulted not significantly different while storage.

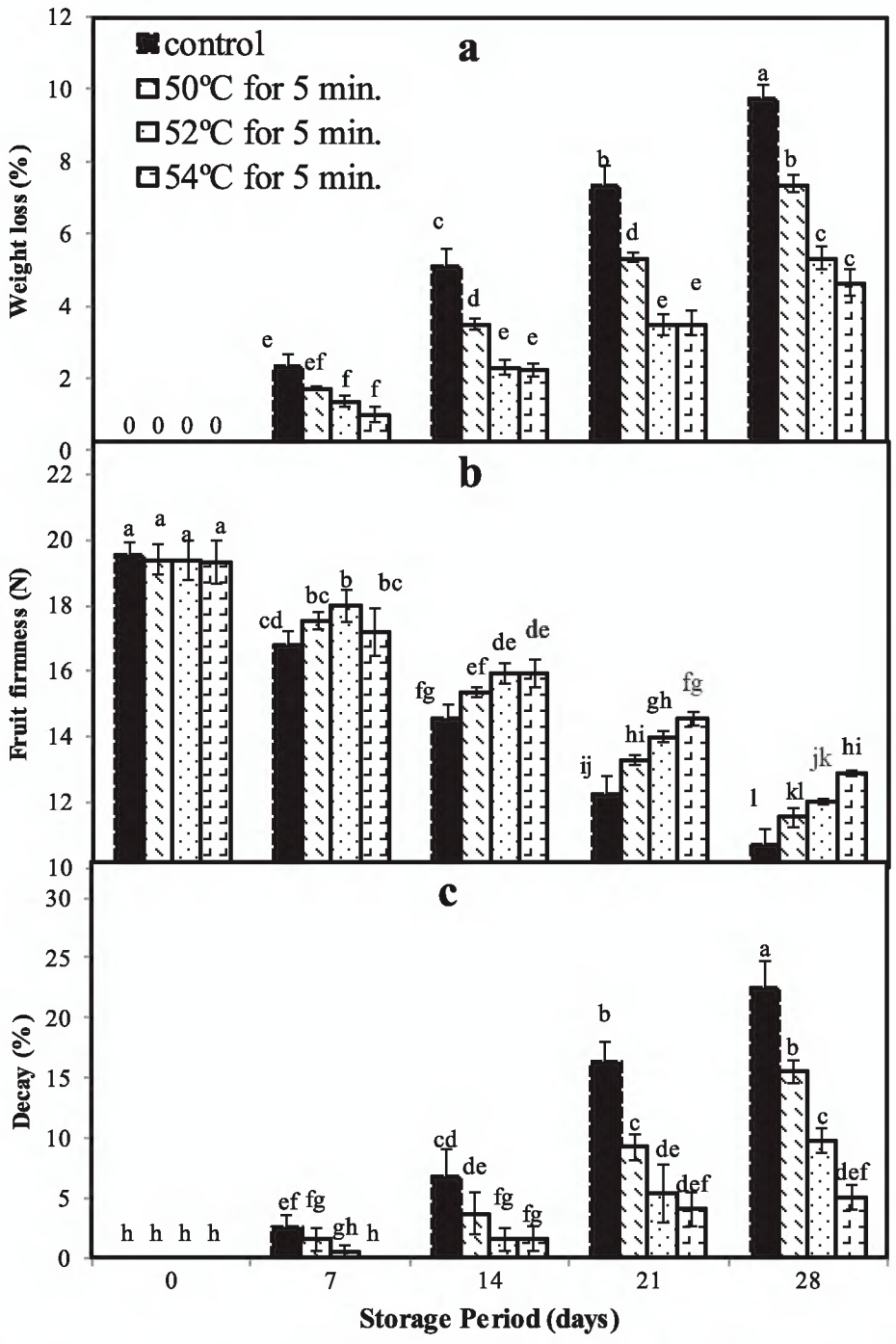

Figure 1. Effect of hot water treatments on the weight loss (**a**), Fruit firmness (**b**) and decaying (**c**) of Red Lady papaya fruit during cold storage for a period of 4 weeks. Data shown are the means of 4 replications. Means sharing same letters are not significantly different from each other at P ≤ 0.05.

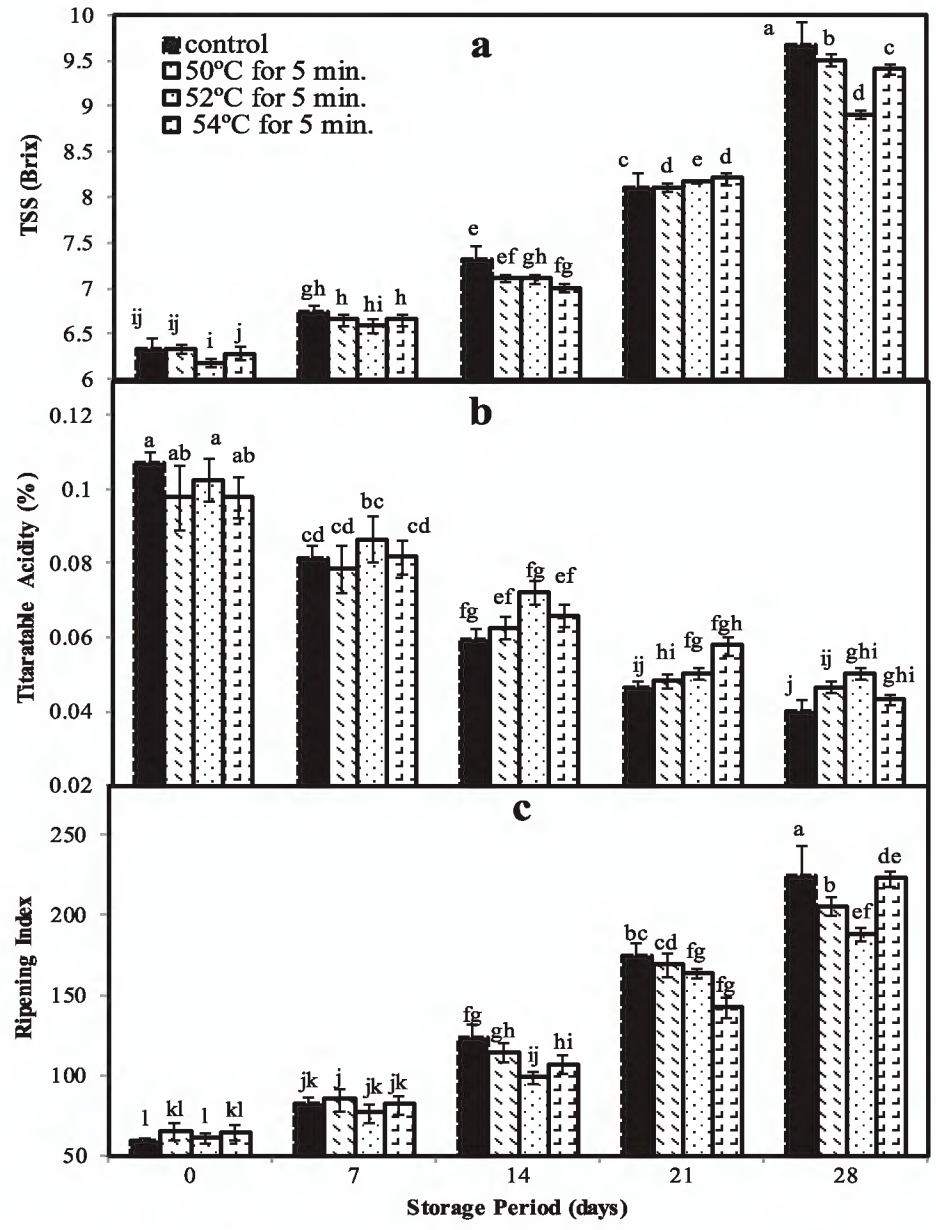

Figure 2. Effect of hot water treatments on total soluble solids (**a**), titratable acidity (**b**) and ripening index (**c**) of Red Lady papaya fruit during cold storage for a period of 4 weeks. The data shown are the means of 4 replications. Means sharing same letters are not significantly different from each other at p ≤ 0.05.

Ripening index is generally TSS/TA in which accumulation of TSS and reduction of TA is considered as ripened index value. Good quality fruit should have a lower ripening index. Fruit at HWT @ 52 °C depicted the minimal index of ripening, while the untreated one showed maximal index of ripening after four weeks. The ripening index of control treated papaya fruit was greater during entire storage, which was 1.6 times more than HWT-treated papaya fruit (Fig. 2c).

Vitamin C, total phenolic content and total antioxidant

The vitamin C contents of papaya fruit were statistically different ($p \leq 0.05$). Maximal vitamin C contents (33 mg/100 g) were present in HWT @ 52 °C- while minimum vitamin C (25 mg/100 g) was noticed in control fruit. Vitamin C contents were lessened with continuation of storage disregarding. Papaya fruits subjected to HWT resulted in lesser reduction in ascorbic acid contents. The vitamin C content of untreated fruit was maximal (34.24 mg/100 g) at 0 day afterward declined till 4th week. It was clearly visible from the entire data that lower temperature for HWT (50 °C) and higher temperature (54 °C) performed at par to each other regarding ascorbic acid content. While fruit exposure to hot water at 52 °C temperature retained maximum level of vitamin C content during entire storage span (Fig. 3a).

Total phenolic contents showed significant variations among HWT treatment and storage periods with their interaction ($p \leq 0.05$). Total phenolic contents of stored papaya fruit were decreased during 1st week of storage then gradually increased till 21st day of storage in all fruit without considering any treatment (Fig. 3b). The total phenolic contents were maximal (35 mg/g FW) in fruit of HWT @ 52 °C and minimal (29 mg/g FW) in fruit of control treatment over the termination of cold storage. Total phenols of papaya fruit both untreated and those treated with lower temperature HWT were minimum initially but reached to the peak level (32 and 33 respectively) on day-21 then declined to the lowest level till the completion of storage spell.

Total antioxidants (%DPPH inhibition) declined along the progress of storage span without considering the any treatments (Fig. 3c). The fruit of all treatment showed reduction in total antioxidant for 0–7 day then increased and again declined during last week of storage. Exposure of fruit to HWT (at 52 °C and 54 °C) showed increase in antioxidants from 7–21 days then declined till 28th day but control fruit and lower temperature of HWT-treated fruit showed increasing trend of antioxidants from 7–14 day then declined later on. Maximum Total Antioxidants (47% DPPH inhibition) were filed in fruit subjected to HWT at 52 °C accompanied by 42% DPPH inhibition, 38% DPPH inhibition and 32% DPPH inhibition in HWT at 54 °C, 50 °C and control respectively. Untreated papaya fruit showed peak level of total antioxidants (%DPPH inhibition) on 14th day while HWT at 52 °C better maintained the antioxidants at upper limit during entire storage of papaya fruit.

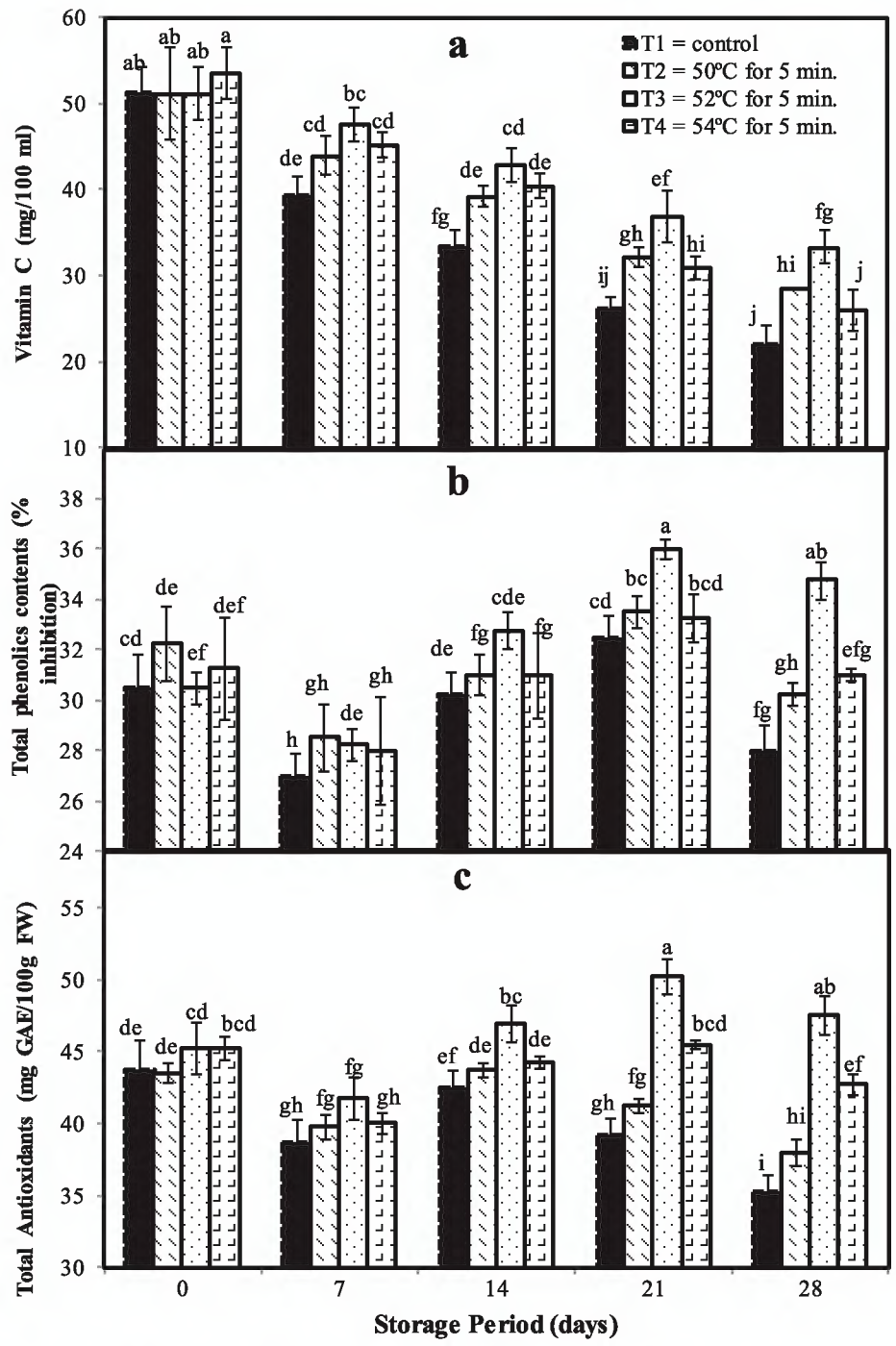

Figure 3. Effect of hot water treatments on vitamin c (**a**), total phenolic contents (**b**) and total antioxidants (**c**) of Red Lady papaya fruit during cold storage for a period of 4 weeks. Data shown are the means of 4 replications. Means sharing same letters are not significantly different from each other at p ≤ 0.05.

Superoxide dismutase, peroxidase and catalase enzymes activities

Superoxide dismutase (SOD) activity exhibited varying patterns with intensified storage duration in consideration of all treatments (Fig. 4a). The fruit of all treatments depicted slight variation in enzyme activity during the first week. Later on, the activity of SOD enzyme was declined in control treatment while inclined in hot water treated papaya fruit. SOD activity of papaya fruit exposed to HWT at 52 °C was maximum on 21st days and declined again during last week of cold storage. SOD activity was maximum (1.1 U/mg protein) in fruit treated with HWT at 52 °C while minimum (0.78 U/mg protein) was in control treatment. Over the time of cold storage period, higher SOD activity (1.41 times more) was noticed in fruit treated with HWT at 52 °C when compared to the control fruit.

Data being statistically analyzed revealed significant results for peroxidase (POD) enzyme activity of papaya fruit treated with HWT. An inverse relation was observed between control treatment and time of storage, depicting that the POD activity was lowered down with progressed storage span. Activity of peroxidase enzyme was decreased in control treatment while increased in fruit subjected to HWT at 52 °C treatment from 0–7 day. From 7–14 days of storage peroxidase activity of all treatments showed increasing trend this again declined till 28th day. After termination of storage maximal POD activity (3.65 U/mg protein) was shown by the fruit subjected to HWT at 52 °C. It was indicated by the results that HWT at 52 °C successfully retained the higher peroxidase activity in papaya fruit under storage (Fig. 4b).

The activity of catalase (CAT) enzyme was remarkably increased by the hot water treatment of papaya fruit (Fig. 4c). During the time of initial 3 weeks of storage the catalase activity was higher while it was lowered down later on in the fruit of control as well as fruit treated with hot water at a temperature of 50 °C. However, fruit treated with HWT at 52 °C showed increasing trend of CAT activity during last week too. Maximal CAT activity (5.6 U/mg protein) was found in HWT at 52 °C treated fruit while minimal (4 U/mg protein) in control fruit on the last day of cold storage (28th).

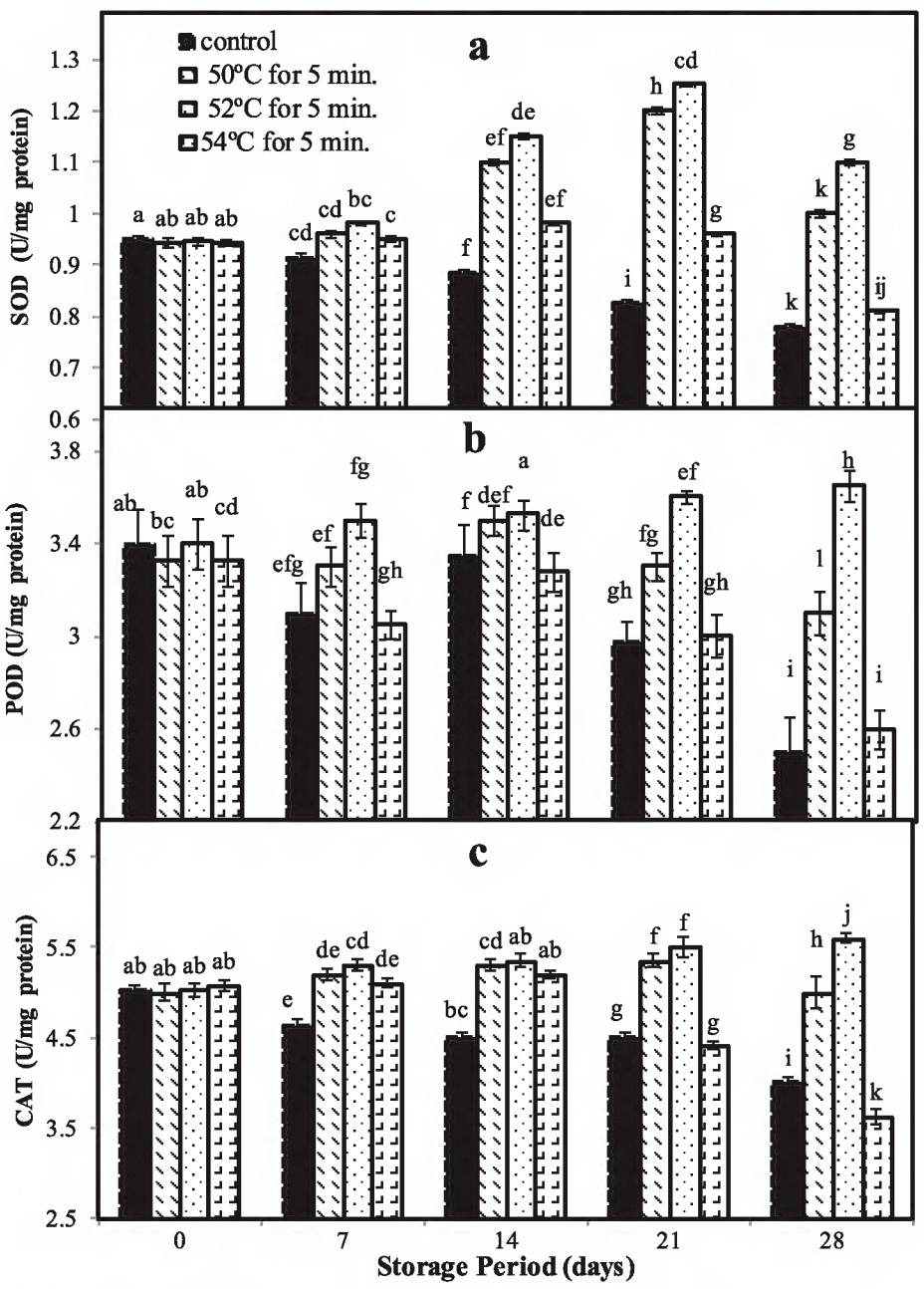

Figure 4. Effect of hot water treatments on SOD (**a**), POD (**b**) and CAT (**c**) enzymes activities of Red lady papaya fruit during cold storage for a period of 4 weeks. Data shown are the means of 4 replications. Means sharing same letters are not significantly different from each other at $p \leq 0.05$.

Discussion

It is really important to maintain the fruit quality and extend storage life of Papaya fruit which is highly perishable and readily affected by various fungal pathogens. Researchers tried different chemical and fungicidal sprays that have been used in past to overcome the problem of decay and storage life with quality, but the use of any chemical is not safe for the Human and environment. Hot water treatment is the best and most effective approach to manage this problem (Fruk et al. 2012; Liu et al. 2012).

In this study, papaya fruit subjected to different hot water levels at pre-storage conditions showed less weight loss with continuation of storage as compared to the control. Continuous respiration in climatic fruit caused reduction in fruit weight by water loss. By using hot water prior to storing the fruit, respiration may be slowed, water loss can be minimized, and the fruit's weight will not decrease as much. Prestorage hot water treatment suppressed the ethylene production, delay ripening and slowed down the rate of respiration in papaya fruit, which might be a reason for less weight loss in response to hot water (Zhao et al. 2008; Rashid et al. 2019).

Increasing the fruit's storage life enhanced its softening, whereas HWT-treated papaya maintained its firmness to a lesser amount. The fact that HWT block the activity of cell wall degrading enzyme and by inhibiting the breakdown of pectic components of the cell wall contributes to the maximum preservation of firmness in response to hot water (Shariah et al. 2013). Pectic substances are an important component of cell wall because they prevent the disassembling of cellulose and hemicellulose (Liu et al. 2009). Prevention of cell wall degradation resulted in more rigidity and firmness. Another factor for less fruit softening may be due to inhibition of ethylene forming substances (Lurie 1998). Fruit firmness reduced in cold storage with passage of time (Mohammad et al. 2011; Fruk et al. 2012).

Papaya fruit infected with fungal pathogens during maturation in the field (Gamagae et al. 2003) and spores persist on the fruit which causes fruit decay during storage. Fungal pathogen can attack in field pre-harvesting and post-harvesting in storage conditioning, but application of hot water treatment in pre-storage inhibits or slows down the fruit decay (Schirra et al. 2000). Application of hot water treatment molts the naturally present waxes on the surface of fruit. Which are then absorbed partially or completely by the stomata present on the surface and the spores present on the skin are inactivated (Fallik et al. 1996). The occupied stomata openings with molten wax and inactivated spores are the major reason for protecting the fruit against decay. Inactivation of initially present spores in response to heat treatment was also reported in fruit (Liu et al. 2012) and filled stomata with waxy material in response to heat treatment has been filed in pear which prevent the fruit from fungal decay.

The biochemical properties of papaya fruit were not affected significantly by hot water treatment during

storage. TSS, acidity and ripening index of stored papaya fruit depicted non-significant variations regarding HWT while significantly varied with expanded storage span. The total soluble solids were enhanced in papaya fruit with passage of time during cold storage and acidity was declined which might be due to utilization of organic acid (acidity) for various metabolic processes undergoing in fruit. Total soluble solids can be related to the loss of water and breakdown of polysaccharides into monosaccharaides. Hot water treatments (50 °C, 55 °C and 60 °C) applied to Satsuma mandarin did not affect the quality of fruit in terms of TSS, TA and their ratio during storage (Hone et al. 2007). Hot water treated (45 °C) mangoes showed non-significant differences regarding biochemical properties (Anwar and Malik 2007). TSS and acidity of papaya fruit exposed to hot water at two different temperatures (42 °C and 49 °C) during ripening was not affected significantly when treated with hot water (Azlin et al. 2013).

Ascorbic acid contents were lessened with continuation of storage in case of all treatments however this reduction was less in hot water treated fruit. The reduction in amount of ascorbic acid could be attributed to its oxidation by ascorbic acid oxidase enzyme catalyzed by polyphenol oxidase (Jiang et al. 2004). It might be possible that HWT suppresses the activity of these enzymes and kept the ascorbic acid contents of stored papaya fruit at upper limit. Promyou et al. (2008) reported that hot water treated two banana cultivars depicted reduced activity of polyphenol oxidase enzyme which catalyzes the destruction of ascorbic acid generally.

Total antioxidants in the present study declined slightly during the first week then increased and finally decreased during last week of storage. The initial decrease may be due to less stress and a strong defense mechanism (as the fruit were unripe). The increase in antioxidant activity might be due to low temperature stress (Suzuki and Mittler 2006). However, HWT also causes mild stress which activates the antioxidant activity of treated commodity to prevent it from further stress (Li 2003). Increased antioxidant activity was also reported in green mum fruit in response to hot water treatment during storage (Endo et al. 2019). Total phenolic contents also exhibited a similar trend like total antioxidant which exhibits their positive correlation with each other. Increase in total phenolic compounds might be attributed to the decreased activity of poly phenol oxidase enzyme which oxidizes total phenolic contents. Promyou et al. (2012) reported that HWT can effectively decrease the activity of poly phenol oxidase enzymes. Decreased activity of phenolic compound oxidizing enzyme in response to hot water treatment was also reported in two banana varieties (Promyou et al. 2008).

In the present study catalase activity of control was declined throughout storage; however HWT significantly increased the catalase activity of papaya fruit during cold storage. Catalase enzyme is one of the important enzymes in the family of antioxidative enzymes. It helps the fruit to scavenge free radicles formed due to oxidative stress (Mittler 2002). When the fruit is exposed to oxidative environment lipid peroxidation of the membrane is increased due to free radicles which ultimately cause fruit softening (Bartoli et al. 1996). Plant cells have their own mechanism to eliminate these free radicles to overcome the damage caused by oxidative stress (Foyer et al. 1994).

Superoxide dismutase is an important enzyme which can destroy the free radicles by converting them into hydrogen peroxide and water (Scandalios 1993). HWT increased the superoxide dismutase level in treated papaya fruit. Heat treatment initiates a signaling pathway in the treated commodity to activate the defense mechanism to defeat further stress. Similar results with increased SOD activity in response to hot water treatment in papaya fruit was reported by Huajaikaew et al. (2005).

Peroxidase activity was increased in papaya fruit treated with HWT as compared to control. Papaya fruit is susceptible to low temperature and shows chilling injury symptoms. Increased level of peroxidase enzyme plays a vital role to overcome cold injury (Wang et al. 2012). Higher activity of POD enzyme might also be related to the over ripening as it is described in past by Wang and Jiao (2001) in blackberries. It was also reported by Singh and Singh (2013) that activities of antioxidative enzymes were enhanced in plum during cold storage.

Conclusion

Hot water treatment was found significantly effective against decay, but fruit treated with HWT @ 52 °C showed less decay, higher antioxidative enzyme activities, maximum total phenolic and total antioxidants during 28 days of cold storage. In conclusion, prestorage treatment of papaya fruit with HWT @ 52 °C for 10 mintues is an effective strategy for suppressing fungal decay and maintaining the good quality of fruit during cold storage.

Statements and declarations

Author contributions

AH, MA, and MA conceptualized the work and conceived the experimental design. MA IH, MA, ZY, and SA collected samples and carried out formal analyses. AH, MAI, KH, STH, SA, and IH performed physiological, biochemical analysis, and statistical analysis. MA, MTA, IM, STH, SA, and MA helped in writing, interpretation of the results and in the revision of the manuscript. All authors have read and agreed to the published version of the manuscript.

Conflict of interest

Authors declare no conflict of interest for this study.

Funding

This research was not funded by any public or private sector organization.

Data availability statement

Any supporting data would be available from Corresponding author upon request.

Acknowledgements

We highly appreciate the support of Mr. Rashid (who provided the premium quality of red lady papaya fruit from his own orchard. We further acknowledged the technical support and kind guidance of Mr. Abdul Rahim Khan from Post-Harvest section of Ayyub Agriculture Research Institute, Faisalabad, Pakistan. This research article has been extracted from the Ph.D. thesis of Aliya Hanif.

References

Ainsworth EA, Gillespie KM (2007) Estimation of total phenolic contents and other oxidation substances in plant tissue using Folin-Ciocalteu reagent. Nature Protocols 2: 875–877. https://doi.org/10.1038/nprot.2007.102

Al-Qurashi AD, Awad MA (2011) Quality characteristics of bisir 'Barhee' dates during cold storage as affected by postharvest dipping in gibberellic acid, naphthaleneacetic acid and benzyladenine. Fruits 66: 343–352. https://doi.org/10.1051/fruits/2011048

Anwar R, Malik AU (2007) Hot water treatment affect ripening quality and storage life of mango. Pakistan Journal of Agricultural Sciences 44(2): 304–311. https://doi.org/10.17660/ActaHortic.2008.768.24

Aravind G, Bhowmik D, Duraivel S, Harish G (2013) Traditional and medicinal uses of *Carica papaya*. Journal of Medicinal Plants Studities 1: 7–15.

Azlin RN, Razal M, Habsah M (2013) Effect of hot water treatment on physicochemical properties of papaya during storage. In: Proc. 7th Interntional Symp. Acta Horticulturae, 479–484. https://doi.org/10.17660/ActaHortic.2013.1012.62

Bartoli CG, Simontacchi M, Montaldi E, Puntarulo S (1996) Oxidative stress, antioxidant capacity and ethylene production during ageing of cut carnation (*Dianthus caryophyllus*) petals. Journal of Experimental Botany 47: 595–601. https://doi.org/10.1093/jxb/47.4.595

Boedeker W, Watts M, Clausing P, Marques E (2020) The global distribution of acute unintentional pesticide poisoning: Estimations based on a systematic review. BMC Public Health 20: 1875. https://doi.org/10.1186/s12889-020-09939-0

Bradford MM (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemisty 72: 248–254. https://doi.org/10.1016/0003-2697(76)90527-3

Brand WW, Cuvellier M, Berset C (1995) Use of a free radical to evaluate antioxidant activity. LWT- Food Science and Technology 28: 25–30. https://doi.org/10.1016/S0023-6438(95)80008-5

Cia P, Pascholati SF, Benato EA, Camili EC, Santos CA (2007) Effects of gamma and UV-C irradiation on the postharvest control of papaya anthracnose. Postharvest Biology and Technology 43: 366–373. https://doi.org/10.1016/j.postharvbio.2006.10.004

Ciou JY, Lin HH, Chiang PY, Wang CC, Charles AL (2011) The role of polyphenol oxidase and peroxidase in the browning of water caltrop pericarp during heat treatment. Food Chemistry 127: 523–527. https://doi.org/10.1016/j.foodchem.2011.01.034

Droby S, Wisniewski M, Macarisin D, Wilson C (2009) Twenty years of postharvest biocontrol research: Is it time for a new paradigm. Postharvest Biology and Technology 52: 137–145. https://doi.org/10.1016/j.postharvbio.2008.11.009

El-Sayyed AI, Mohamed AH, Odero DC, Goma AM (2019) Biochemical effects of chitosan coating and hot water dipping on green bean rot during cold storage. Journal of Applied Sciences 19(2): 101–108. https://doi.org/10.3923/jas.2019.101.108

Endo H, Ose K, Bai J, Imahori Y (2019) Effect of hot water treatment on chilling injury incidence and antioxidative response of mature green mume fruit during low temperature storage. Scientia Horticulturae 246: 550–556. https://doi.org/10.1016/j.scienta.2018.11.015

Fallik E (2004) Prestorage hot water treatments (immersion, rinsing and brushing). Postharvest Biology and Technology 32: 125–134. https://doi.org/10.1016/j.postharvbio.2003.10.005

Fallik E, Grinberg S, Alkalai S, Lurie S (1996) The effectiveness of postharvest hot water dipping on the control of grey and black moulds in sweet red pepper (*Capsicum annuum*). Plant Pathology 45: 644–649. https://doi.org/10.1046/j.1365-3059.1996.d01-175.x

Foyer C, Descourvieres P, Kunert K (1994) Protection against oxygen radicals: An important defence mechanism studied in transgenic plants. Plant Cell Environment 17: 507–523. https://doi.org/10.1111/j.1365-3040.1994.tb00146.x

Fruk G, Nišević L, Sever Z, Miličević T, Jemrić T (2012) Effects of different postharvest heat treatments on decreasing rot, reducing chilling injury and maintaining quality of nectarine fruit. Agriculturae Conspectus Scientificus 77: 27–30. https://hrcak.srce.hr/77899

Gamagae S, Sivakumar D, Wijesundera R (2004) Evaluation of post-harvest application of sodium bicarbonate-incorporated wax formulation and candida oleophila for the control of anthracnose of papaya. Journal of Crop Protection 23: 575–579. https://doi.org/10.1016/j.cropro.2003.11.003

Gamagae S, Sivakumar D, Wijeratnam RW, Wijesundera R (2003) Use of sodium bicarbonate and candida oleophila to control anthracnose in papaya during storage. Journal of Crop Protection 22: 775–779. https://doi.org/10.1016/S0261-2194(03)00046-2

Hanif A, Ahmad S, Jaskani MJ, Ahmad R (2020a) Papaya treatment with putrescine maintained the overall quality and promoted the antioxidative enzyme activities of the stored fruit. Scientia Horticulturae 268: 109367. https://doi.org/10.1016/j.scienta.2020.109367

Hanif A, Ahmad S, Shahzad S, Liaquat M, Anwar R (2020b) Postharvest application of salicylic acid reduced decay and enhanced storage life of papaya fruit during cold storage. Journal of Food Measurement and Characterization 14: 3078–3088. https://doi.org/10.1007/s11694-020-00555-5

Hernández Y, Lobo MG, González M (2006) Determination of vitamin C in tropical fruit: A comparative evaluation of methods. Food Chemistry 96: 654–664. https://doi.org/10.1016/j.foodchem.2005.04.012

Hodges DM, Lester GE, Munro KD, Toivonen PM (2004) Oxidative stress: Importance for postharvest quality. HortScience 39: 924–929. https://doi.org/10.21273/HORTSCI.39.5.924

Hong SI, Lee HH, Kim D (2007) Effects of hot water treatment on the storage stability of satsuma mandarin as a postharvest rot control. Postharvest Biology and Technology 43: 271–279. https://doi.org/10.1016/j.postharvbio.2006.09.008

Horrigan L, Lawrence RS, Walker P (2002) How sustainable agriculture can address the environment and human health harms of industrial agriculture. Environmental Health Perspectivest 110(5): 445–456. https://doi.org/10.1289/ehp.02110445

Hortwitz W (1960) Official and tentative methods of analysis, 9th Ed. Association of Official Agricultural Chemists. Washington, D.C, 314–320.

Huajaikaew L, Uthairatanakij A, Kanlayanarat S, Gemma H (2005) Effect of heat treatment on antioxidants in papaya fruit stored at low temperature. Acta Horticulturae 6822: 1063–1068. https://doi.org/10.17660/ActaHortic.2005.682.139

Jemric T, Ivic D, Fruk G, Matijas HS, Cvjetkovic B, Bupic M, Pavkovic B (2011) Reduction of postharvest rot of peach and nectarine caused by monilinia laxa using hot water dipping. Food Bioprocess Technology 4: 149–154. https://doi.org/10.1007/s11947-010-0355-z

Jiang Y, Duan X, Joyce D, Zhang Z, Li J (2004) Advances in understanding of enzymatic browning in harbested litchi fruit. Food Chemistry 88: 443–448. https://doi.org/10.1016/j.foodchem.2004.02.004

Jouki M, Khazaei N (2014) Effect of low-dose gamma radiation and active equilibrium modified atmosphere packaging on shelf life extension of fresh strawberry fruits. Food Packaging and Shelf Life 1(1): 49–55. https://doi.org/10.1016/j.fpsl.2013.12.001

Li MH (2003) Peroxidase and superoxide dismutase activities in Fig leaves in response to ambient air pollution in a subtropical city. Archives of Environmental Contamination. Toxicology 45: 168–176. https://doi.org/10.1007/s00244-003-0154-x

Liu D, Zou J, Meng Q, Zou J, Jiang W (2009) Uptake and accumulation and oxidative stress in garlic (*Allium sativum* L.) under lead phytotoxicity. Ecotoxicology 18: 134–143. https://doi.org/10.1007/s10646-008-0266-1

Liu J, Sui Y, Wisniewski M, Droby S, Tian S, Norelli J, Hershkovitz V (2012) Effect of heat treatment on inhibition of monilinia fructicola and induction of disease resistance in peach fruit. Postharvest Biology Technology 65: 61–68. https://doi.org/10.1016/j.postharvbio.2011.11.002

Lu X, Sun D, Mo Y, Xi J, Sun G (2010) Effects of post-harvest salicylic acid treatment on fruit quality and anti-oxidant metabolism in pineapple during cold storage. Journal of Horticulture Science and Biotechnology 85: 454–458. https://doi.org/10.1080/14620316.2010.11512697

Lurie S (1998) Postharvest heat treatments. Postharvest Biology Technology 14: 257–269. https://doi.org/10.1016/S0925-5214(98)00045-3

Mari M, Torres R, Casalini L, Lamarca N, Mandrin JF, Lichou J, Larena I, De Cal MA, Melgarejo P, Usall J (2007) Control of post-harvest brown rot on nectarine by *Epicoccum nigrum* and physico-chemical treatments. Journal of the Science of Food and Agriculture 87: 1271–1277. https://doi.org/10.1002/jsfa.2839

Mittler R (2002) Oxidative stress, antioxidants and stress tolerance. Trends Plant Sciences 7: 405–410. https://doi.org/10.1016/S1360-1385(02)02312-9

Nunes M, Emond J, Brecht J (2006) Brief deviations from set point temperatures during normal airport handling operations negatively affect the quality of papaya (*Carica papaya*) fruit. Postharvest Biology Technology 41: 328–340. https://doi.org/10.1016/j.postharvbio.2006.04.013

Oloyede O (2005) Chemical profile of unripe pulp of *Carica papaya*. Pakistan Journal of Nutrition 4: 379–381. https://doi.org/10.3923/pjn.2005.379.381

Promyou S, Ketsa S, Doorn WG (2008) Hot water treatments delay cold-induced banana peel blackening. Postharvest Biology Technology 48: 132–138. https://doi.org/10.1016/j.postharvbio.2007.09.006

Promyou S, Supapvanich S, Boodkord B, Thangapiradeekajorn M (2012) Alleviation of chilling injury in jujube fruit (*Ziziphus jujuba* Mill) by dipping in 35 C water. Kasetsart Journal Nature Science 46: 107–119. http://kasetsartjournal.ku.ac.th/kuj_files/2012/A1203081448285817.pdf

Rashid HA, Barman BC, Hasna MK, Begum HA (2019) Effect of non chemical treatment on postharvest diseases, shelf life and quality of papaya under two different maturity stages. Journal Bangladesh Agricultural University 17(1): 14–25. https://doi.org/10.3329/jbau.v17i1.40658

Rui H, Cao S, Shang H, Jin P, Wang K, Zheng Y (2010) Effects of heat treatment on internal browning and membrane fatty acid in loquat fruit in response to chilling stress. Journal of Science Food and Agriculture 90: 1557–1561. https://doi.org/10.1002/jsfa.3993

Scandalios J (1993) Oxygen stress and superoxide dismutase. Plant Physiology 101: 7–12. https://doi.org/10.1104/pp.101.1.7

Schirra M, D'hallewin G (1997) Storage performance of fortune mandarins following hot water dips. Postharvest Biology Technology 10: 229–238. https://doi.org/10.1016/S0925-5214(96)01301-4

Schirra M, Hallewin GD, Inglese P, La Mantia T (1999) Epicuticular changes and storage potential of cactus pear [*Opuntia ficus-indica* Miller (L.)] fruit following gibberellic acid preharvest sprays and postharvest heat treatment. Postharvest Biology Technology 17: 79–88. https://doi.org/10.1016/S0925-5214(99)00043-5

Schirra M, D'hallewin G, Ben-Yehoshua S, Fallik E (2000) Host–pathogen interactions modulated by heat treatment. Postharvest Biology and Technology 21: 71–85. https://doi.org/10.1016/S0925-5214(00)00166-6

Schirra M, Mulas M, Fadda A, Mignani I, Lurie S (2005) Chemical and quality traits of 'Olinda' and 'Campbell' oranges after heat treatment at 44 or 46°C for fruit fly disinfestation. LWT-Food Science and Technolgy 38: 519–527. https://doi.org/10.1016/j.lwt.2004.07.011

Shalom NB, Hanzon J, Pinto R, Lurie S (1996) Cell wall changes and partial prevention of fruit softening in prestorage heat treated 'Anna' apples. Journal of the Science of Food and Agriculture 72: 231–234. https://doi.org/10.1002/(SICI)1097-0010(199610)72:2%3C231::AID-JSFA644%3E3.3.CO;2-1

Shariah SN, Luqman ZAM, Mahmud TMM, Ahmad SH (2013) Hot water dips affects shelf life and postharvest quality of kendondong fruit. 7th Proc. Int. Post. Symp. Acta Horticulturae, 539–546. https://doi.org/10.17660/ActaHortic.2013.1012.72

Singh SP, Singh Z (2013) Dynamics of enzymatic and non-enzymatic antioxidants in japanese plums during storage at safe and lethal

temperatures. LWT-Food Science Technology 50: 562–568. https://doi.org/10.1016/j.lwt.2012.08.008

Steel RGD, Torrie JH, Dicky DA (1997) Principles and Procedures of Statistics: A Biometrical Approach. 3rd Edition, McGraw Hill, Inc. Book Co., New York.

Suzuki N, Mittler N (2006) Reactive oxygen species and temperature stresses: A delicate balance between signaling and destruction. Physiologia Plantarum 126: 45–51. https://doi.org/10.1111/j.0031-9317.2005.00582.x

Villa-Rojas R, López-Malo A, Sosa-Morales ME (2011) Hot water bath treatments assisted by microwave energy to delay postharvest ripening and decay in strawberries (*Fragaria* × *ananassa*). Journal of the Science of Food and Agriculture 91(12): 2265–2270. https://doi.org/10.1002/jsfa.4449

Wang H, Zhang Z, Xu L, Huang X, Pang X (2012) The effect of delay between heat treatment and cold storage on alleviation of chilling injury in banana fruit. Journal of the Science Food and Agriculture. 92: 2624–2629. https://doi.org/10.1002/jsfa.5676

Zhao N, Li X, Chen W, Shi J (2008) Effect of hot water treatment on the inhibition of anthracnose, PG, PME activity and PGIP gene expression in harvested papaya fruit. 4th Int Symp. Tropical and Subtropical Fruits, 975 pp. https://doi.org/10.17660/ActaHortic.2013.975.63